



## ORIGINAL ARTICLE

# Sublingual Estradiol Offers No Apparent Advantage Over Combined Oral Estradiol and Cyproterone Acetate for Gender-Affirming Hormone Therapy of Treatment-Naive Trans Women: Results of a Prospective Pilot Study

Iris Yaish,<sup>1,†</sup> Guy Gindis,<sup>1,2,†</sup> Yona Greenman,<sup>1,2</sup> Yaffa Moshe,<sup>1</sup> Mira Arbiv,<sup>1</sup> Assaf Buch,<sup>1,2</sup> Yael Sofer,<sup>1,2</sup> Gabi Shefer,<sup>1</sup> and Karen Tordjman<sup>1,2,\*</sup>

### Abstract

**Purpose:** Chronic gender-affirming hormone therapy (GAHT) with sublingual estradiol (SLE) has not been studied. We aimed to compare GAHT with SLE only, to combined oral (CO) estradiol and cyproterone acetate, in treatment-naive trans women.

**Methods:** Twenty-two trans women enrolled into either the CO arm or the SLE-only arm (0.5 mg four times daily) in this 6-month prospective study. Anthropometric and laboratory variables were collected at baseline and 3 and 6 months. At the study beginning and end, body composition was measured by dual-energy X-ray absorptiometry and bioelectrical impedance, and gender dysphoria, sexual desire, and function were assessed by validated questionnaires.

**Results:** Subjects in the SLE were older,  $26.3 \pm 5.8$  years versus  $20.1 \pm 2.3$  years,  $p = 0.006$ . All anthropometric, body composition, and laboratory variables were identical at baseline. Although dysphoria appeared greater, and sexual function lower at baseline in the CO group, this canceled out after age adjustment. Both treatments induced similar biochemical and hormonal changes. Creatinine, hemoglobin and cholesterol decreased significantly, while testosterone was suppressed to the same level in both groups:  $3.22 [1.47-5.0]$  nmol/L in the SLE group and  $2.41 [0.55-8.5]$  nmol/L in the CO,  $p = 0.65$ . Significant changes in body composition toward a more feminine body were noted in both groups. Dysphoria did not significantly improve in either group, while sexual desire and function decreased at six months in both,  $p < 0.001$ .

**Conclusions:** Both treatments achieved similar clinical changes. At this stage, SLE, which repeatedly induces alarming excursions of serum estradiol throughout the day, appears to offer no advantage over the CO approach.

**Keywords:** cyproterone acetate; gender dysphoria; gender-affirming hormone therapy; nonbinary; sublingual estradiol; transgender women

### Introduction

Gender-diverse people are coming out in ever-growing numbers.<sup>1</sup> Among them, many gender-fluid and non-binary people also request gender-affirming hormonal therapy (GAHT). Most current standards of care

address the needs of nonbinary people, but fall short of offering specific guidance, and simply reiterate general recommendations.<sup>2-4</sup> This perceived lack of expertise is sending transgender people searching for GAHT protocols that might accomplish what they are seeking,

<sup>1</sup>Institute of Endocrinology, Metabolism, and Hypertension, Tel Aviv Sourasky Medical Center, Tel Aviv, Israel.

<sup>2</sup>Faculty of Medicine, Tel Aviv University, Tel Aviv-Yafo, Israel.

<sup>†</sup>Joint first co-authorship, both authors contributed equally to this work.

\*Address correspondence to: Karen Tordjman, MD, Institute of Endocrinology, Metabolism, and Hypertension, Tel Aviv-Sourasky Medical Center, 6 Weizmann Street, Tel Aviv 6423906, Israel, E-mail: karent@tlvmc.gov.il

often glean information from informal sources such as internet forums and other web-based platforms.<sup>5</sup>

In our experience, a growing number of trans women, particularly nonbinary subjects, prefer not to take an antiandrogen, and switch to a sublingual estradiol (SLE)-only protocol, based on the unproven assumption that it might better preserve libido and erectile function.<sup>6</sup>

Although the possibility of a feminizing protocol based on the administration of estradiol only is suggested in the literature,<sup>7–10</sup> the efficacy and safety of chronic SLE have never been assessed.<sup>11</sup> As demonstrated in earlier studies of post-menopausal women,<sup>12</sup> and more recently in trans women,<sup>13</sup> the pharmacokinetics of SLE differ from those of a similar orally ingested dose. While oral estradiol peaks at 6–8 h, with a  $T_{1/2}$  of up to 20 h, SLE peaks after 1–2 h, and results in serum estradiol concentrations that are at least four to five times higher than those achieved by the oral route.<sup>13</sup> Moreover, due to the rapid waning of estradiol after sublingual administration, there is a need for repeated administration throughout the day to maintain therapeutic levels.

In this study, we sought to assess the efficacy of chronic SLE-only GAHT of treatment-naïve, trans women, and to compare this approach to the conventional combined estradiol and cyproterone acetate (CPA) protocol (combined oral [CO]) practiced at our institution. Based on preliminary data, we anticipated to see higher testosterone concentrations, and better preservation of sexual desire/function among the subjects receiving SLE.

## Methods

### Study design

This was a 6-month, prospective, controlled, open-label and nonrandomized study of treatment-naïve, trans women who sought GAHT at the Transgender Health Center within the institute of Endocrinology, Metabolism and Hypertension, at Tel Aviv-Sourasky Medical Center, between November 2021 and June 2022.

### Protocol

Healthy, adult (18 years of age and older), treatment-naïve, trans women seeking GAHT were invited to participate in the study. The nonrandomized, open-label design of this pilot study was intended to improve compliance, especially with the sublingual arm, which required four times daily administration of the hormone. Smokers, subjects with a history of attempt-

ed suicide, drug or alcohol abuse, or significant hypertriglyceridemia (fasting triglycerides  $\geq 400$  mg/dL), a personal history of malignancy in the preceding 5 years, a family history of thrombophilia, or evidence of primary hypogonadism were excluded. After giving written informed consent and receiving explanations regarding the protocol, each subject stated her study arm preference.

The CO protocol consisted of once-daily 2 mg estradiol as hemihydrate (Estrofem; Novo Nordisk), and 10 mg CPA (Androcur; Bayer), while the estradiol-only protocol (SLE) consisted of 0.5 mg of estradiol taken sublingually, four times a day, roughly every 6 h.

### Measures

At baseline, every subject underwent a complete physical examination, and laboratory assessment of biochemical (Advia Centaur XP autoanalyzer; Siemens), hematological, and hormonal variables in the fasting state, between 08:00 and 09:00, making sure the subject had been awake at least 2 h before sampling. Estradiol, luteinizing hormone (LH), follicle-stimulating hormone (FSH), prolactin, and prostate-specific antigen were determined by chemiluminescence (Immulite<sup>®</sup> 2000; Siemens), and testosterone by electro-chemiluminescence (COBAS 6000, e 691 module; Roche).

Body composition was analyzed by dual-energy X-ray absorptiometry (Lunar Prodigy 11; GE Healthcare, Chicago, IL), and by bioelectrical impedance (InBody770; InBody, Cerritos, CA). Finally, each subject was requested to respond to three validated questionnaires: The gender congruence and life satisfaction scale (GCLS) to assess gender dysphoria;<sup>14</sup> the sexual desire inventory (SDI)<sup>15,16</sup>; and the international index of erectile function (IIEF).<sup>17,18</sup>

All laboratory measurements were repeated at 3 and 6 months. For subjects in the SLE arm, serum estradiol level was also measured 90 min after taking the morning dose to get a fuller assessment of the peak levels these subjects were exposed to. This step was omitted for subjects in the CO arm as oral estradiol peaks between 6 and 8 h, and the waiting time to catch this level would have been impractical. Body composition measurements and questionnaires were again administered, and a breast Tanner stage was determined upon completion of the study at 6 months.

### Sample size calculation

Preliminary data (Supplementary Table S1), suggested that testosterone concentrations would be higher at 6

months in subjects in the SLE arm. Taking into account a possible “lost to follow-up” rate of 30%, with a power of 80% and an alpha value of 0.05, we calculated that 11 participants would be needed in each arm to demonstrate the primary end-point, a testosterone concentration  $\geq 66\%$  higher in subjects in the sublingual arm, compared to that measured with the control treatment.

### Statistical methods

Continuous variables were inspected for normality and expressed either as mean  $\pm$  standard deviation or median with interquartile range. For continuous variables, comparisons between groups were made either by Student's *t*-test or by the Mann–Whitney tests. Paired comparisons were performed with the paired *t*-test, or the Wilcoxon ranked-sign test. Frequencies were assessed by the Fisher's exact test or the chi-square test. Univariate correlations between the scores on the questionnaires and other continuous variables were analyzed by the Pearson or Spearman tests. Linear regressions were performed to adjust for independent variables on the questionnaires. Statistical computations were performed with SPSS version 28.0 (IBM Corp., Armonk, NY). Statistical significance was assumed for  $p < 0.05$ .

### Ethical considerations

The study protocol was approved by the institutional human studies ethics committee (approval no. TLV-21-0325).

## Results

### Baseline data

Although this was not a randomized study, the major difference at baseline between the groups was age, as subjects opting for the SLE-only protocol were older than those who chose the CO treatment,  $26.3 \pm 5.8$  years versus  $20.1 \pm 2.3$  years,  $p = 0.006$  (Table 1). The youngest participant was 18 years and the oldest 39 years. Selected variables are included in Table 1; all other baseline biochemical and anthropometric results are found in the Supplementary Table S2.

Baseline dysphoria and sexual desire/function measures. At baseline, the groups differed in the GCLS score. Subjects who opted for the CO protocol had higher levels of dysphoria than subjects who chose SLE,  $112 \pm 15.3$  versus  $92.9 \pm 15.1$ ,  $p = 0.009$ . Despite a nonsignificantly lower score on the SDI,  $58.5 \pm 24.4$  in the CO group versus  $74.4 \pm 17.5$  in the

**Table 1. Baseline Demographic, Selected Anthropometric, and Biochemical Data of Study Subjects**

| Variable                 | SLE, <i>n</i> = 11  | CO, <i>N</i> = 11   | <i>p</i> |
|--------------------------|---------------------|---------------------|----------|
| Age (year)               | 26.3 (5.8)          | 20.1 (2.3)          | 0.006    |
| Binary/nonbinary (N/n)   | 8/3                 | 9/2                 | 0.5      |
| Married/single (N/n)     | 2/9                 | 0/11                | 0.24     |
| BMI (kg/m <sup>2</sup> ) | 25.1 [20.7–27.4]    | 22.5 [20.9–29.5]    | 0.75     |
| WHR                      | 0.883 [0.831–0.914] | 0.833 [0.804–0.967] | 0.61     |
| Body fat (%)             | 25.3 [18.7–30.4]    | 22.7 [12.4–33.2]    | 0.95     |
| Creatinine (mg/dL)       | 0.99 [0.83–1.03]    | 0.9 [0.78–1.0]      | 0.37     |
| Hematocrit (%)           | 44.2 [42.0–47.1]    | 45.5 [44.0–47.5]    | 0.44     |
| Testosterone (nmol/L)    | 18.6 [15.4–21.4]    | 17.2 [14.5–27.4]    | 0.85     |
| Bioavailable T (nmol/L)  | 7.8 [5.3–10.5]      | 8.6 [5.5–10.3]      | 0.92     |
| LH (IU/L)                | 4.6 [3.6–5.8]       | 3.5 [3.0–4.5]       | 0.33     |
| FSH (IU/L)               | 4.0 [3.6–5.0]       | 2.8 [2.1–4.4]       | 0.12     |
| Estradiol (pmo/L)        | 117 [90.3–145]      | 112 [92.6–142]      | 0.85     |
| Prolactin (mIU/L)        | 163 [142–234]       | 179 [143–270]       | 0.52     |

Data are presented as mean with SD, or median with IQR.

BMI, body mass index; CO, combined oral; FSH, follicle-stimulating hormone; IQR, interquartile range; LH, luteinizing hormone; SD, standard deviation; SLE, sublingual estradiol; T, testosterone; WHR, waist-to-hip ratio.

SLE one,  $p = 0.094$ , the results on the IIEF sexual function questionnaire were significantly lower in these subjects  $32.5 \pm 16.7$  versus  $48.7 \pm 15.1$ ,  $p = 0.027$ . However, baseline dysphoria intensity was negatively correlated with age (Pearson  $r = -0.447$ ,  $p = 0.037$ ); therefore, when adjusted for age, the difference between the treatment groups was no longer significant. Likewise, after age adjustment, the baseline IIEF score at baseline between the groups was no longer significantly different.

### Post-treatment results

Although repeat laboratory measurements were obtained at 3 months (Supplementary Table S2), the following will focus on the study outcomes at 6 months.

Effect of GAHT on laboratory variables. Both treatments induced expected biochemical and hematological changes compared to baseline, but there was no difference between the treatment groups. Not surprisingly, in parallel to the decrease in lean body mass (see effect of GAHT on body composition), serum creatinine decreased from  $0.91 \pm 0.13$  to  $0.76 \pm 0.12$  mg/dL at 6 months,  $p < 0.001$ . Total cholesterol also decreased from a median of 161.5 [139–179.5] to 137 [133.5–175.5] mg/dL,  $p = 0.013$ , low-density lipoprotein cholesterol decreased as well from a median of 89.1 [78.4–118.5] to 86 [69–108] mg/dL,  $p = 0.008$ . High-density lipoprotein cholesterol and triglycerides were unaltered. We observed a significant decrease in serum total bilirubin

and alkaline phosphatase, while other liver function tests were unchanged in both treatment groups (see Table 2 for selected data at baseline and 6 months, and Supplementary Table S2 for all data).

Also expected under treatment, for the entire cohort, as both groups did not differ in this respect neither at baseline nor at 6 months, we observed a significant decrease both in hemoglobin concentration, from  $15.2 \pm 0.9$  to  $13.9 \pm 0.95$  g/dL, and hematocrit, from  $45.2\% \pm 2.5\%$  to  $40.6\% \pm 3.0\%$ ,  $p < 0.001$ , for both paired comparisons, but no change in basic coagulation parameters (Supplementary Table S2).

**Effect of GAHT on sex hormones.** Contrarily to our expectations, both treatments decreased testosterone profoundly and we found no difference between the treatment groups (Fig. 1). At 6 months, the median testosterone in the SLE group was  $3.22$  [1.47–5.0] nmol/L, while it was  $2.41$  [0.55–8.5] nmol/L in the CO treatment group,  $p = 0.65$ . Surprisingly, despite an equal daily dose, the oral estradiol treatment was less efficient in raising basal morning estradiol concentrations, which remained unchanged compared to baseline

(Fig. 1). In contrast, the sublingual administration led to an almost doubling of basal/trough estradiol concentrations at 6 months, such that these were significantly higher in the SLE group,  $204.5 \pm 63.3$  pmol/L versus  $109.7 \pm 53$  pmol/L,  $p = 0.002$ .

Interestingly, although SLE did not result in significantly higher testosterone than the CO treatment at 6 months, LH concentrations decreased significantly less with this protocol than with the CO regimen (Fig. 1). Indeed, LH concentrations in these subjects were  $4.5 \pm 1.3$  IU/L versus  $1.6 \pm 1.3$  IU/L,  $p = 0.004$ . In contrast, FSH decreased significantly in the SLE group only (Fig. 1). However, this did not result in a significant difference between the groups at 6 months,  $0.39$  [0.24–1.55] IU/L in the SLE group versus  $1.57$  [0.59–2.35] IU/L in the CO group,  $P = \text{NS}$ .

CPA is known to be the main culprit for the prolactin elevation seen with GAHT in trans women.<sup>19,20</sup> Indeed, by paired comparisons, prolactin increased significantly only in the CO group, from  $179$  [143–270], to  $350.4$  [241–765] mIU/L,  $p < 0.005$ .

Finally, subjects in the SLE arm underwent measurements of serum estradiol 90 min after taking the morning 0.5 mg dose. This dose generated an increase in estradiol from a basal value of  $204.5 \pm 63.5$  to a peak of  $1994 \pm 1457$  pmol/L (range 229–4736),  $p = 0.001$ .

**Table 2. Selected Baseline and 6-Month Biochemical and Hematological Variables, Presented for the Entire Cohort, as There Was No Difference in Any of the Variables Between the Treatment Groups**

| Variable                                         | Baseline, $n = 22$ | 6 Months, $n = 21$ | $p$    |
|--------------------------------------------------|--------------------|--------------------|--------|
| Glucose (mg/dL)                                  | 87.5 (7.1)         | 83.1 (15.7)        | 0.08   |
| Creatinine (mg/dL)                               | 0.91 (0.13)        | 0.76 (0.12)        | <0.001 |
| ALT (U/L)                                        | 23 [19.8–26]       | 19 [17–31.5]       | 0.31   |
| AST (U/L)                                        | 18 [13.5–25.3]     | 19 [13–25]         | 0.76   |
| Alkaline phosphatase (U/L)                       | 79.7 (28.9)        | 62 (14.5)          | <0.001 |
| Bilirubin (mg/dL)                                | 0.69 (0.27)        | 0.53 (0.19)        | 0.04   |
| Cholesterol (mg/dL)                              | 161.5 [139–179.5]  | 137 [133.5–175.5]  | 0.01   |
| HDL-cholesterol (mg/dL)                          | 50 [40.8–53]       | 48 [39.7–59.5]     | 0.68   |
| LDL-cholesterol (mg/dL)                          | 89.1 [78.4–118.5]  | 86 [68–108]        | 0.008  |
| Triglycerides (mg/dL)                            | 89.5 [60.5–134]    | 84 [62.5–98.5]     | 0.113  |
| Hemoglobin (g/dL)                                | 15.2 (0.9)         | 13.9 (0.95)        | <0.001 |
| Hematocrit (%)                                   | 45.2 (2.5)         | 40.6 (3.0)         | <0.001 |
| Red blood cells ( $10^6/\text{mm}^3$ )           | 5.29 [5.03–5.52]   | 4.66 [4.4–4.9]     | <0.001 |
| White blood cells (cell $\times 10^9/\text{L}$ ) | 6.62 (1.36)        | 7.35 (1.38)        | 0.04   |
| Platelets (cells $\times 10^9/\text{L}$ )        | 249.5 (57.2)       | 256.8 (69.4)       | 0.22   |
| INR                                              | 1.01 (0.06)        | 1.0 (0.05)         | 0.39   |
| TSH (mIU/L)                                      | 1.93 [1.27–2.4]    | 1.83 [1.52–2.56]   | 0.78   |

Data are given as mean with SD, or median with IQR, as indicated. Paired comparisons were done accordingly, either by the paired  $t$ -test or the Wilcoxon matched-pairs signed rank test.

ALT, alanine transaminase; AST, aspartate transaminase; HDL, high-density lipoprotein; INR, international standardized ratio; LDL, low-density lipoprotein; TSH, thyroid-stimulating hormone.

**Effect of GAHT on body composition.** Trans women are usually desirous to achieve more feminine body proportions, and this was indeed achieved with both treatments. Although body mass index was unchanged at 6 months in either group, both treatments induced significant changes at 6 months as the percentage of body fat increased from  $24.0 \pm 10.4$  to  $28.8 \pm 8.5$ ,  $p < 0.001$ , by paired  $t$ -test. Likewise, the percentage of gynoid fat increased from  $28.7 \pm 10.1$  to  $33.3 \pm 7.4$  after 6 months,  $p = 0.002$ . This resulted in a significant decrease in the android/gynoid fat ratio from  $1.05 \pm 0.18$  to  $0.93 \pm 0.18$ ,  $p < 0.001$ , and in an equally significant decrease in the waist-to-hip ratio (WHR), from  $0.883 \pm 0.08$  (median: 0.88; [0.78–0.93]) to  $0.86 \pm 0.07$  (median: 0.85; [0.81–0.86]),  $p = 0.009$  by Wilcoxon.

These changes were echoed by measurements of absolute body compartment masses. Total fat mass increased from  $17,566$  [10,652–25,394] to  $18,598$  [14,585–25,128] g,  $p = 0.007$ ; and lean body mass decreased from  $51,906 \pm 6121$  to  $48,707 \pm 6503$  g,  $p < 0.001$ . Of note, there was no difference in any of these variables between treatments.

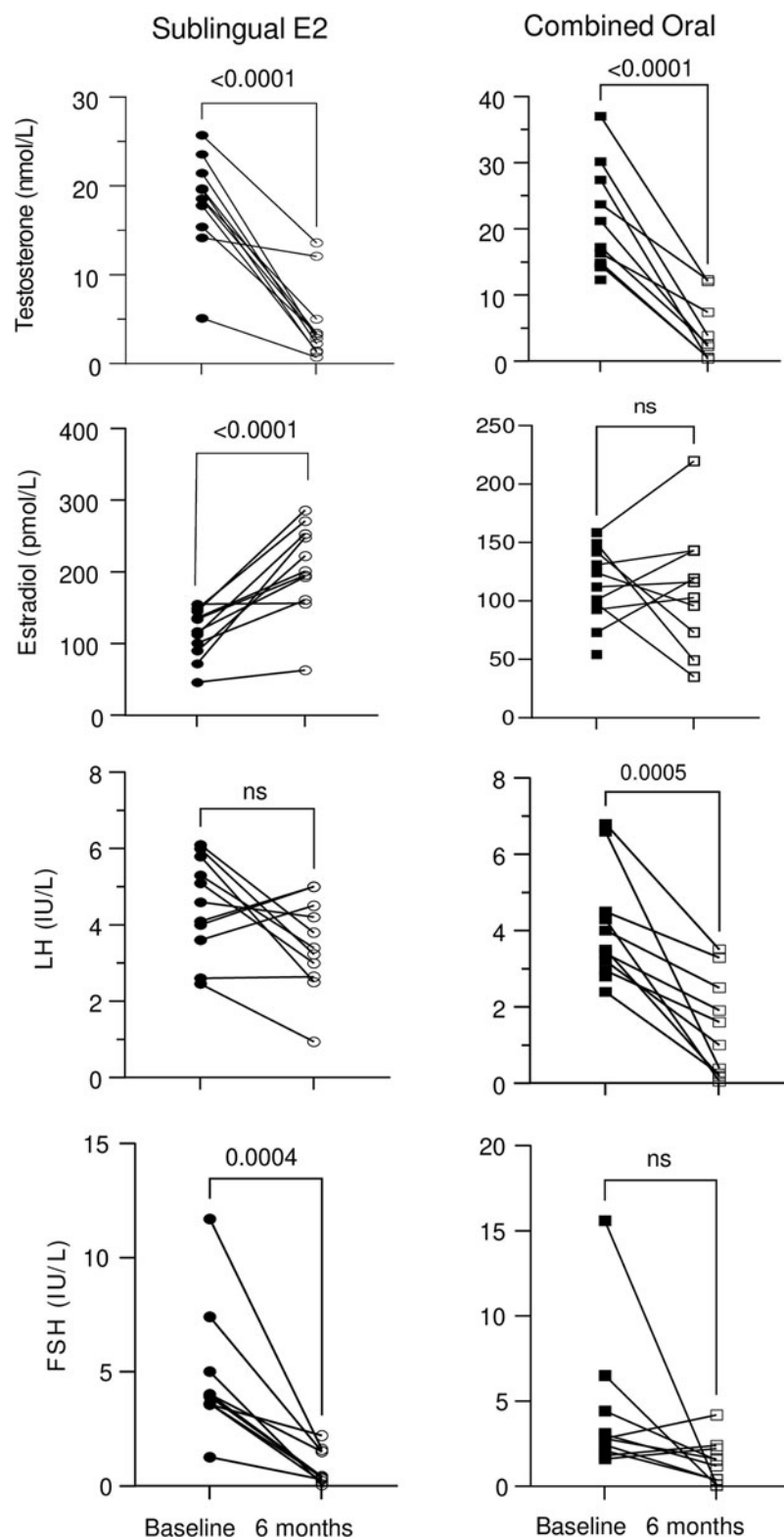


**FIG. 1.** Hormonal changes under sublingual estradiol only, or the combined oral treatment in treatment-naïve trans women. Paired comparisons by *t*-test or Wilcoxon matched-pairs signed rank test were performed between baseline (filled symbols) and 6 months (empty symbols). All values were from blood samples taken fasting, before the morning dose. E2, estradiol; FSH, follicle-stimulating hormone; LH, luteinizing hormone.

Finally, breast development was indistinguishable between the treatment groups; the most frequent stage, reached in both arms, was Tanner 3.

**Effect of GAHT on dysphoria and sexuality.** We analyzed the scores on all three questionnaires after 6 months of treatment compared to baseline, both in absolute and relative terms (raw data are found in Supplementary Tables S3–S5).

**GCLS measure of gender dysphoria.** Over the 6 months that the study lasted, there was no convincing improvement in the GCLS as assessed by paired *t*-test in either treatment group. For the entire cohort, baseline dysphoria score was  $102.3 \pm 17.7$  and  $100 \pm 13.3$  at 6 months,  $p = 0.7$  (maximal possible score = 266, a higher score denoting worse dysphoria). There was, however, a very significant negative correlation between the baseline dysphoria score and the extent of its reduction by 6 months, Pearson correlation  $r = 0.63$ ,  $p = 0.002$ . Namely, the more severe the dysphoria at baseline, the smaller its improvement after 6 months of GAHT (computed as the absolute difference between the baseline score and that at 6 months).

Indeed, in a multivariate linear regression, the only determinant of dysphoria at 6 months was the intensity of dysphoria at baseline ( $\beta = 0.606$ ,  $p < 0.001$ ), while age, testosterone level, and treatment group did not contribute to this measure.

**SDI measure of sexual desire.** GAHT induced a significant reduction in sexual desire compared to baseline in both groups,  $p < 0.001$ , for all subjects. Although there was a significant difference in the SDI score between the groups at 6 months,  $37.9 \pm 23.5$  in the CO group versus  $58.5 \pm 16$  in the SLE,  $p = 0.03$  (maximal possible score = 126), the percent decrease relative to baseline was similar,  $-26.5\% \pm 45.9\%$  versus  $-19.6\% \pm 19.2\%$ , respectively,  $p = 0.65$ . Again, the only determinant of the 6-month sexual desire score was the baseline score ( $\beta = 0.586$ ,  $p = 0.006$ ). Age and treatment group had no independent contribution.

**IIEF measure of sexual function.** Here too, GAHT caused a significant attenuation in sexual and erectile function in both treatment groups, from  $40.6 \pm 17.6$  to  $34.1 \pm 16.2$  (maximal possible score = 70),  $p < 0.001$ , by paired comparisons for the entire cohort. However, the individual percent change was similar between groups,  $-25.7\%$  [ $-47.8$ – $18$ ] in the CO treatment

group versus  $-16.2\%$  [ $-30$ – $10.4$ ] for subjects in the SLE group,  $p = 0.56$ . Again, the only predictor of sexual function at 6 months was sexual function at baseline ( $\beta = 0.697$ ,  $p < 0.001$ ).

Notably, neither sexual desire nor sexual function was in any way correlated with testosterone concentration at 6 months.

## Discussion

This pilot study is, to the best of our knowledge, the first evaluation of the efficacy of chronic SLE for GAHT of treatment-naïve trans women. We embarked on this study under the assumption that sublingual treatment without an antiandrogen would suppress testosterone less than CO and spare some of the consequent decline in sexual desire and function associated with treatment.

This assumption, which permeates the transgender internet-based culture,<sup>21,22</sup> was not supported by our findings.

Despite some significant interindividual responses in both treatment groups, testosterone concentrations were suppressed to the same degree by both GAHT approaches. Interestingly, LH levels were less suppressed by the SLE approach, suggesting CPA was responsible for the more profound suppression of LH in the CO treatment arm. Our data also reinforced the notion that it is predominantly CPA that is responsible for the prolactin elevation in the CO protocol.<sup>19,20,23</sup>

Given the rapid clearance of estradiol after a sublingual administration,<sup>12,13</sup> the lower trough estradiol levels in the CO group were a surprise. As orally ingested estradiol has been shown to have a prolonged half-life, this could be the result of the fairly low dose of estradiol used in this study.<sup>24</sup> This study was not intended to provide information regarding estradiol fluctuations throughout the day, but the extreme excursions in estradiol concentration documented 90 min after the sublingual administration should be kept in mind when recommending sublingual estrogen to trans women.

Over the limited study period, both treatments appeared similar from the biochemical and hematological standpoints, generally indistinguishable in the changes they generated, and by and large consistent with previous reports.<sup>25–28</sup> Although this study did not go into an extensive assessment of thrombogenic factors, there was no signal of any derangement, and no relevant clinical adverse effect.

That GAHT rapidly achieved the desired changes in body composition toward a more feminine shape was

rewarding. Indeed, we observed an early decrease in lean body mass, an increase in fat mass and in total body fat percentage, and a change toward a more female fat distribution, including a decrease in both the android/gynoid fat ratio and WHR. All these have been previously reported<sup>29–31</sup>; however, with the exception of a small Korean study,<sup>32</sup> these changes were never documented in trans women earlier than 12 months into treatment. Importantly, we show that both treatments are equally effective in bringing about those early changes.

This was a short study in which one of the outcomes we attempted to assess was the intensity of dysphoria after initiation of GAHT. The generally held view is that GAHT improves gender dysphoria, as well as dysphoria-related psychological conditions, such as anxiety and depression.<sup>33</sup> However, a precise assessment of how dysphoria evolves with GAHT is difficult to obtain due to the multitude of instruments to gauge it, and less than perfect agreement between them.<sup>34</sup> Most prospective studies that indicate a clear improvement in this outcome have assessed it after several years of treatment (from 2 to 8), often in combination with gender-affirming surgery.<sup>35,36</sup> Recently, a short-term prospective study showed a statistically significant, but clinically questionable, improvement in dysphoria in 30 trans women after 6 months of GAHT, using a 12-item questionnaire based on a 2-week recall.<sup>37</sup>

For this study, we chose to use the GCLS. This validated 38-item questionnaire, interrogates the responder over the preceding 6 months.<sup>14,38</sup> We thus felt it would be an appropriate reflection of the study duration. Although still to be proven, using the GCLS tool in a longer study might have revealed an absolute improvement in dysphoria. For now, all we observed was a negative correlation between baseline dysphoria severity and its decrease at 6 months, independent of the treatment approach.

The decrease in sexual desire and sexual function we observed under both treatment protocols came as no surprise. Although we had anticipated to see preserved sexual desire and function in the SLE group, given the same documented decrease in testosterone in both groups, this outcome was completely consistent. Decrease in sexual desire is an expected early consequence of feminizing GAHT, and usually recovers with the passing of time.<sup>16</sup> Again, a later evaluation of study subjects might have shed a definite answer to this question.

### Study limitations

Although this was a prospective controlled study, the nonrandom treatment allocation introduced a significant age bias, which explained most of the baseline differences between the groups (dysphoria and sexual desire and function). Nowadays when access to treatment is less a barrier, it is conceivable that transgender people who present at an older age suffer from a lesser degree of dysphoria, and have had greater opportunities to experiment with sexual activity. In addition, the sample size was small and was based on an assumption that proved incorrect.

Finally, the study period was limited, and although on almost all the laboratory variables, there were no differences between 3 and 6 months, there is a possibility that a longer study duration could have unraveled convincing improvement in dysphoria, and further changes in body composition.

Nevertheless, the study also has some real strengths. First and foremost, its novelty, as no study had previously shown results of chronic GAHT with SLE. Furthermore, despite its small sample size and short duration, it was exhaustive. Among its noteworthy results, the lack of apparent superiority of the sublingual route, and the suggestion that an antiandrogen may not be needed to achieve adequate testosterone suppression.

### Conclusions

In this first comparative study of GAHT in treatment-naïve transgender women, SLE alone offered no real advantage over the standard CO estradiol and CPA protocol. Both approaches yielded tangible and similar results.

Finally, multiple daily dosing of SLE is less convenient. Moreover, this generates recurring exceedingly high serum estradiol concentrations, the thrombogenicity of which remains to be evaluated.

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### Authors' Contributions

I.Y.: Conceptualization, investigation, supervision, and article reviewing and editing. G.G.: Investigation, resources, and article reviewing and editing. Y.G.: Conceptualization and article reviewing and editing. Y.M.: Investigation. M.A.: Investigation. A.B.: Conceptualization, supervision, and article reviewing and editing. Y.S.: Conceptualization and article reviewing and

editing. G.S.: Investigation, resources, and article reviewing and editing. K.T.: Conceptualization, investigation, supervision, formal analysis, original draft writing, and article reviewing and editing.

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### Supplementary Material

Supplementary Table S1

Supplementary Table S2

Supplementary Table S3

Supplementary Table S4

Supplementary Table S5

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### Abbreviations Used

ALT = alanine transaminase  
 AST = aspartate transaminase  
 BMI = body mass index  
 CO = combined oral  
 CPA = cyproterone acetate  
 FSH = follicle-stimulating hormone  
 GAHT = gender-affirming hormone therapy  
 GCLS = gender congruence and life satisfaction scale  
 HDL = high-density lipoprotein  
 IIEF = international index of erectile function  
 INR = international standardized ratio  
 IQR = interquartile range  
 LDL = low-density lipoprotein  
 LH = luteinizing hormone  
 SD = standard deviation  
 SDI = sexual desire inventory  
 SLE = sublingual estradiol  
 T = testosterone  
 TSH = thyroid-stimulating hormone  
 WHR = waist-to-hip ratio